

CLAIMS

1. A method of recycling a solid support for cultivation of anchorage-dependent cells located within a system for cell cultivation comprising the steps of:
  - 5 a) emptying said system of liquid;
  - b) rinsing said system with an aqueous solution ;
  - c) rinsing said system with a sodium hydroxide solution; and
  - d) rinsing said system with an aqueous solution.
2. The method of claim 1, wherein said aqueous solution is water.
- 10 3. The method of claim 1 or 2, wherein said sodium hydroxide solution is at a concentration selected from the group consisting of:
  - a) within a range of about 1% to about 3% sodium hydroxide;
  - b) within a range of about 1.5 % to about 2.5 % sodium hydroxide; and
  - c) about 2% sodium hydroxide.
- 15 4. The method of any of claims 1 to 3, wherein step (d) is performed at least three times.
5. The method of claim 4, wherein step (d) is performed three times.
6. The method of claim 4, wherein step (d) is performed five times.
7. The method of any of claims 1 to 6, wherein said aqueous solution in step (b) is Water For Injection (WFI).
- 20 8. The method of any of claims 1 to 7, wherein said aqueous solution in step (d) is Water For Injection (WFI).
9. The method of any of claims 4 to 7, wherein step (d) is performed with Purified Water (PW) except for the last repetition of step (d), which is performed with Water For Injection (WFI).
- 25 10. The method of any of claims 1 to 9, wherein said solid support is made of non -woven fibrous matrix bonded to a porous support sheet.
11. The method of claim 10, wherein said solid support is a disk made of non -woven polyester bonded to a sheet of polypropylene mesh.
12. The method of any of claims 1 to 9, wherein said solid support is a microcarrier.
- 30 13. The method of any of claims 1 to 12, wherein said system comprises a bioreactor.
14. The method of claim 13, wherein said system further comprises an external column connected to said bioreactor.

15. The method of claim 13, wherein said bioreactor comprises an internal column.
16. The method of any of claims 14 or 15, wherein said solid support is located within said column.
17. The method of any of claims 1 to 16, wherein said system is sterilized after performing step (d).
18. The method of claim 14 or 16, wherein said external column is sterilized after performing step (d).
19. The method of any of claims 1 to 18, wherein step (d) is carried out with a circulation loop flow selected from the group consisting of:
  - 10 a) within a range of about 500  $\text{L.h}^{-1}.\text{kg}^{-1}$  to about 700  $\text{L.h}^{-1}.\text{kg}^{-1}$ ;
  - b) within a range of about 550  $\text{L.h}^{-1}.\text{kg}^{-1}$  to about 650  $\text{L.h}^{-1}.\text{kg}^{-1}$ ; and
  - c) about 583  $\text{L.h}^{-1}.\text{kg}^{-1}$ .
20. The method of any of claims 1 to 19, wherein step (d) is carried out at ambient temperature.
- 15 21. The method of any of claims 1 to 20, wherein step (d) is carried out under an overpressure selected from the group consisting of:
  - a) within a range of about 100 millibars to about 900 millibars;
  - b) within a range of about 300 millibars to about 700 millibars; and
  - c) about 500 millibars.
- 20 22. The method of any of claims 1 to 21, wherein step (d) is carried out for a duration selected from group of:
  - a) within a range of about 5 minutes to about 30 minutes;
  - b) within a range of about 5 minutes to about 20 minutes; and
  - c) about 10 minutes.
- 25 23. The method of any of claims 1 to 22, wherein step (c) is carried out with a circulation loop flow set at a value selected from the group consisting of:
  - a) within a range of about 500  $\text{L.h}^{-1}.\text{kg}^{-1}$  to about 700  $\text{L.h}^{-1}.\text{kg}^{-1}$ ;
  - b) within a range of about 550  $\text{L.h}^{-1}.\text{kg}^{-1}$  to about 650  $\text{L.h}^{-1}.\text{kg}^{-1}$ ; and
  - c) about 583  $\text{L.h}^{-1}.\text{kg}^{-1}$ .
- 30 24. The method of any of claims 1 to 23, wherein step (c) is carried out for a duration selected from group of:
  - a) within a range of about 20 minutes to about 40 minutes;
  - b) within a range of about 25 minutes to about 35 minutes; and
  - c) about 30 minutes.

25. The method of any of claims 1 to 24, wherein step (c) is carried out at a temperature selected from the group consisting of:

5           a) within a range of about 50°C to about 70 minutes;  
          b) within a range of about 55°C to about 65°C minutes; and  
          c) about 60°C.

26. The method of any of claims 1 to 25, wherein step (b) is carried out at a temperature selected from the group consisting of:

10           a) within a range of about 50°C to about 70°C;  
          b) within a range of about 55°C to about 65°C; and  
          c) about 60°C.

27. The method of any of claims 1 to 26, wherein step (b) is carried out under an overpressure selected from the group consisting of:

15           a) within a range of about 100 millibars to about 900 millibars;  
          b) within a range of about 300 millibars to about 700 millibars; and  
          c) about 500 millibars.

28. The method of any of claims 1 to 27, wherein step (b) is carried out for a duration selected from group of:

20           a) within a range of about 5 minutes to about 30 minutes;  
          b) within a range of about 5 minutes to about 20 minutes; and  
          c) about 10 minutes.

29. A solid support for cultivation of anchorage-dependent cells recycled according to the method of any of claim 1 to 28.

30. The solid support of claim 29, wherein said solid support is a disk made of non-woven fibers bonded to a porous support sheet.

31. The method of claim 30, wherein said solid support is a disk made of non-woven polyester bonded to a sheet of polypropylene.

32. The solid support of claim 29, wherein said solid support is a microcarrier.

33. Use of the solid support of any of claim 29 to 32 for cultivation of cells.

30           34. The use of claim 33, wherein said solid support is made of non-woven fibers bonded to a porous support sheet.

35. The use of claim 34, wherein said solid support is a disk made of non-woven polyester bonded to a sheet of polypropylene.

36. The use of claim 33, wherein said solid support is a microcarrier.
37. The use of any of claims 33 to 36, wherein said cell is anchorage-dependent.